

Rule 126
F1
cont
(a) generating a three dimensional model of an alpha-amylase structure, utilizing data from Appendix 1 and a computer programmed for generating said model from said data;

(b) identifying in said three-dimensional alpha-amylase structure generated in step (a) at least one amino acid residue or at least one structural part; wherein an alteration in said at least one amino acid residue or said at least one structural part is predicted to result in an altered property, and wherein said altered property is selected from the group consisting of substrate specificity, substrate binding, substrate cleavage pattern, temperature stability, pH dependence of enzymatic activity, pH dependence of stability, stability towards oxidation, Ca^{2+} -dependency and specific activity;

(c) modifying the sequence of a nucleic acid encoding said parent alpha-amylase to produce a nucleic acid encoding a deletion, insertion, or substitution of one or more amino acids at a position corresponding to said at least one amino acid residue or at least one structural part identified in step (b); and

(d) expressing the modified nucleic acid of step (c) in a host cell to produce said variant alpha amylase.

82
80 (new) The method according to claim *81*, which further comprises in step (c) using modeling methods.

83
81 (new) The method according to claim *81*, wherein said three-dimensional alpha amylase structure has an A domain, a B domain and a C domain, wherein said A domain has an amino acid sequence corresponding to residues 1-103 and 206-305 of SEQ ID NO: 2; said B domain has an amino acid sequence corresponding to residues 104-205 of SEQ ID No. 2 and said C domain has an amino acid sequence corresponding to residues 396-483 of SEQ ID NO.2.

but 62

84-82 (new) A method for producing a variant of a parent alpha-amylase having an altered property relative to said parent alpha-amylase, wherein said altered property is selected from the group consisting of substrate specificity, substrate binding, substrate cleavage pattern, temperature stability, pH dependence of enzymatic activity, pH dependence of stability, stability towards oxidation, Ca^{2+} -dependency and specific activity, wherein said parent alpha-amylase has a sequence of at least 70% homology to the sequence of SEQ ID No: 13, when homology is determined by the GAP program (Genetic Computer Group, Version 7.3) using default values for GAP penalties, said method comprising

(a) generating a model of a three dimensional structure of an alpha-amylase structure, defined by the atomic coordinates shown in Appendix 1;

Cont (b) utilizing said model generated in step (a) and modeling methods to identify at least one amino acid residue or at least one structural part; wherein an alteration in said at least one amino acid residue or said at least one structural part is predicted to result in an altered property, and wherein said altered property is selected from the group consisting of substrate specificity, substrate binding, substrate cleavage pattern, temperature stability, pH dependence of enzymatic activity, pH dependence of stability, stability towards oxidation, Ca^{2+} -dependency and specific activity;

(c) modifying the sequence of a nucleic acid encoding said parent alpha-amylase to produce a nucleic acid encoding a deletion, insertion, or substitution of one or more amino acids at a position corresponding to said at least one amino acid residue or at least one structural part identified in step (b); and

(d) expressing the modified nucleic acid of step (c) in a host cell to produce said variant alpha amylase.

85-83 (new) The method according to claim 82 in which step (a) further comprises using a computer programmed for generating said model

Sub G3 86
(new) A method for producing a variant of a parent alpha-amylase having an altered property relative to said parent alpha-amylase, wherein said altered property is selected from the group consisting of substrate specificity, substrate binding, substrate cleavage pattern, temperature stability, pH dependence of enzymatic activity, pH dependence of stability, stability towards oxidation, Ca^{2+} -dependency and specific activity, wherein said parent alpha-amylase has a sequence of at least 70% homology to the sequence of SEQ ID No: 13, when homology is determined by the GAP program (Genetic Computer Group, Version 7.3) using default values for GAP penalties, said method comprising

F1
(a) generating a model of a three dimensional structure of an alpha-amylase structure using a computer programmed for generating a model structure and atomic coordinates shown in Appendix 1;

(b) utilizing said model generated in step (a) and modeling methods to identify at least one amino acid residue or at least one structural part; wherein an alteration in said at least one amino acid residue or said at least one structural part is predicted to result in an altered property, and wherein said altered property is selected from the group consisting of substrate specificity, substrate binding, substrate cleavage pattern, temperature stability, pH dependence of enzymatic activity, pH dependence of stability, stability towards oxidation, Ca^{2+} -dependency and specific activity;

(c) modifying the sequence of a nucleic acid encoding said parent alpha-amylase to produce a nucleic acid encoding a deletion, insertion, or substitution of one or more amino acids at a position corresponding to said at least one amino acid residue or at least one structural part identified in step (b); and

(d) expressing the modified nucleic acid of step (c) in a host cell to produce said variant alpha amylase.